



BIOREMOVAL OF LEAD BY SOME FUNGI ISOLATED FROM SOIL

Fadhil Sami Zghair^{1,*} and Muna Sabbar Jebar²

^{1*}Al-Kufa Technical Institute, Al-Furat Al-Awsat Technical University, Kufa, Iraq.

²Babylon Technical Institute, Al-Furat Al-Awsat Technical University, Babylon, Iraq.

Abstract

Eco-friendly biological removal of heavy metal pollution from liquid factories wastes or soil is a recent approach which using the biosorbent. It has lots of advantages over chemical approach like its low cost, more efficient, more effective, multiple uses etc. The fungi are one of the most desirable absorbents because of its cell wall characteristics. The present work, therefore, aimed to use *Aspergillus niger*, *Alternaria alternata* and *Trichoderma harzianum* isolated from lead-contaminated soil to remove lead from solution. The aim of this work was to study the resistance and removal of lead metal by the fungi. Soil sample collected from contaminated area by lead in Hilla city. Its tolerance lead metal ions was studied by growing fungus in different metal concentration. Bio absorption of lead by live fungal biomass was determined by Atomic absorption spectrophotometer. The results showed that *A.niger* and *A.alternata* grew at 1,800 ppm of lead, while *T.harzianum* grew at 1,400 ppm. With regard to lead removal potential, this removal was achieved 4.9 mg / dL for *A.niger* at a concentration of 600 ppm and 6.2 mg / dL for *A. alternata* at a concentration of 200 ppm and 5.02 mg / dL for *T.harzianum* at a concentration of 1000 ppm In a million As well Different absorption at other concentrations.

Key words: Lead pollution, biomass of fungus, biosorption, *Aspergillus niger*, *Alternaria alternata* and *Trichoderma harzianum*

Introduction

Bio absorption is the removal of substances from the solution of biological materials, the substances may be organic or inorganic and the soluble or insoluble form. Eco-friendly biological removal of heavy metal pollution from liquid factories wastes or soil is a recent approach which using the biosorbent (Shumate and Stranberg, 1985). It has lots of advantages over chemical approach like its low cost, more efficient, more effective, multiple uses etc (Abedin, 2014). The fungi are one of the most desirable absorbents because of its cell wall characteristics. Detoxification by increase the uptake of pollutants by microorganisms (Swati, *et al.*, 2019). Recently, increase in population number and the massive develop in the industrial revolution increases pollution through accumulation of pollutants such as (Copper, Lead, Cadmium, Nickel and Tin) from industry, Cars, etc. which causes a big problem to environment by altering micro biome and increasing the incidence of diseases (Fawzy, *et al.*, 2017). Microorganisms play a critical role in elimination of pollution, therefore, recent studies focused on treating this matter by using Fungi. Fungi have been

used widely in different aspects such as production of antibiotics, enzymes, alcohol, food products, detoxification, and elimination of industrial wastes (Zaidi and Pal, 2017). The focusing on the use of biological alternatives become very important for that result in a lower alteration of the environment, specifically through the use of microorganisms for the removal of heavy metals or biosorption (Fawzy, *et al.*, 2017). Fungi are ubiquitous members of subaerial and subsoil environments, and often become a dominant grouping metal-rich or metal-polluted habitats (.Moctezuma Zarate, *et al.*, 2017). Recent studies have shown that the strains isolated from contaminated areas have remarkable potential to tolerate such toxic conditions. Microorganisms have been shown to possess ability to survive by adapting or mutating at high concentrations of heavy metals (Yang *et al.*, 2009). Therefore this project aimed for removal of lead ion by a locale fungal strain which is highly resistant to some heavy metals.

Materials and Methods

Fungi Isolation and Identification

Soil sample collected from contaminated area by lead

*Author for correspondence : E-mail : zghair.fa@gmail.com

in Hilla city for isolated the fungi from the polluted soil by using the culture media Potato Dextrose Agar (PDA). 10g. soil was suspended in 90 ml sterile water then serially diluted and plated on PDA media, incubation at 25°C and purification of fungal colonies and identification according the morphological characterizes by classification keys of Pitt and Hocking (1997) and Ellis *et al.*, (2007).

Determination of MIC of Lead

MIC, of lead for fungal isolates were determined a plate method (Ajjabi and Chouba, 2009), were provided by $(\text{CH}_3\text{COO})_2\text{Pb}$ at varying concentration (200-1800 ppm) and 0.5 cm disc from fungal isolates colony was incubated at 25°C for 7 days, and the growth of the plates was compared with a control.

Obtaining the Fungal Biomass

Biosorption of lead by pretreated biomass of the fungi is also used for biosorption of lead with modification of its biomass using the physical and chemical method.

The fungus was grown in large 500 mL flasks containing 250 mL PDA medium. Inoculums (3 disc 0.5 cm, 3 days old cultures in same the medium) was added to the medium and flasks were then kept into the shaking incubator at 100 rpm for 21 days. After this incubation, Biomass samples were rinsed three times with distilled water. Samples were prepared by digestion of 0.5g of dried biomass with 5 mL of conc. HNO_3 and 1.5mL of conc. HCl and by further dilution with Milli-Q water to a final volume of 10 mL. Putted in oven at 90C for two hours complete the value to 10 ml recorded the result by atomic absorption spectrophotometer (Ivan, *et al.*, 2014).

Statistical Analysis

Data are expressed as the mean \pm standard error of measurements from triplicate experiments. One-way analysis of variance (ANOVA) was used to test the significance of differences among the absorbed Pb concentrations, using STATISTICA software, version 5.0 (StatSoft, Inc.). *P* values less than 0.01 were considered significant.

Results and Discussion

Fungi Isolation and Identification

In the present study, a total of three soil fungi were isolated from the heavy metal contaminated soils. Based on the morphological characters, the fungal isolates were identified as *A. niger*, *A. alternata* and *T. harzianum* according to the Pitt and Hocking (1997) and Ellis *et al.*, (2007).

Determination of MIC of Lead

The results show in table 1 that *A. niger* more than

Table 1: Effect of Lead on fungi isolates growth.

Lead con.	Colony diameter Fungi cm		
	<i>A.niger</i>	<i>A. alternata</i>	<i>T.harzianum</i>
0	9	9	9
200	9	9	8.5
600	8.3	8.1	7.3
1000	6.1	5.7	4.1
1400	4.4	3.8	1.5
1800	2.7	2.2	0

other fungi at 1800 ppm lead metal by average colony growth was 2.7 cm diameter, while *A. alternata* 2.2 cm and *T. harzianum* no growth at the same concentration.

Also, other concentrations differed in inhibiting growth of fungal colonies, according to the concentration value and fungus table 1 proved the theory that various genera and also isolates of the same genus do not necessarily have the same heavy metal tolerance (Bejoysekhar, 2015). It comes into mind that tolerance skill is not inherited among microorganisms, in other word it is acquired from ecosystem (Rodriguez *et al.*, 2018).

Biosorption of lead by live and pretreated biomass of fungi

The results are shown in Fig. 1. The fungi removed efficiently most of the lead metal analyzed. *A.alternata* is the best isolate for removal lead was 6.2 mg/dl at con. 200 ppm while *T.harzianum* is removed 5.2 mg/dl at con. 1000 ppm and *A.niger* is removed 4.9 mg/dl at con.600 mg/dl. Also the ability to absorb lead was decreased with increasing concentration and different isolates.

The fungus used in the present study was isolated from lead-contaminated industrial effluent (Mahish, *et al.*, 2015). *A. niger*, *A. alternata* and *T. harzianum* were isolated from soil having the high concentration of lead was able to absorb the lead metal ions. Thus, the fungi isolated from metal contaminated sited were more efficient to tolerate and absorb the heavy metals simply due to the adaptation (Anahid, *et al.*, 2011) and (Siokwu and Anyanwn, 2012). All these strains has found among the

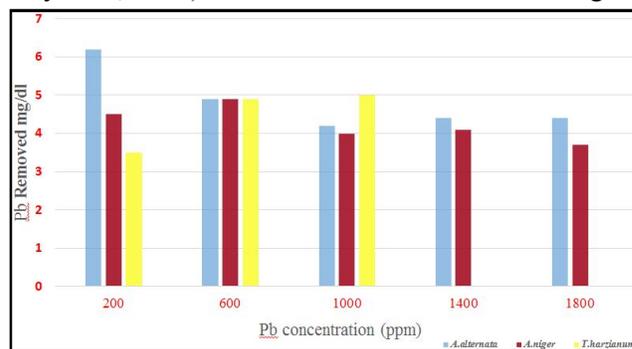


Fig. 1: The pb removed by some fungal isolates (*A. alternata*, *A. niger* and *T. harzianum*) after 21 days incubation.

other fungi with highest lead effectively removal from lead contaminated soils. Lead to a toxic compound which existed in inorganic and organic forms. Different strains have different ability in bio-indicators of lead pollution and able to sporulate well on the lead exposure as compared to control and other strains.

Some most recent research study also supporting present study in view of this genes were used for removal of various heavy metals from polluted water. As some study indicated the possibility of using *A. niger* to remove lead and did not indicated to use *A. alternata* and *T. hazianum* to remove lead ion.

This result has proven that the filamentous fungi isolated from soil contaminated lead sites have more tolerance strains fungi and it can be exploited for removing lead ions from aqueous solution.

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